

Evaluation of New Antibodies by Immunoperoxidase

Procedure for Post evaluation (performed by immunohistochemistry technologist).

Materials

1. Several known positive staining tissues selected by the Director of Immunohistology.
2. Several negative staining tissues.
3. Using vendor's recommendations, a test protocol is entered on new antibody evaluation form. For example, pretreatment (high or low pH retrieval, digestion, etc.), digestion or no pretreatment.
4. Cut positive control tissue that was selected by Director or Designee. This may include a list obtained from vendor specification sheet.
5. Set up for a concentrated test run using several dilutions and recommended pretreatments. For example, 1:10, 1:1:50, 1:100, 1:200, 1:300 after antigen retrieval or digestion. Dilute antibody with PBS. For predilute antibodies, test using different retrieval solutions as well as Protease Digestion.
6. Label all slides with antibody, pretreatment, dilution and date.
7. Proceed with IHC Automated Procedure
8. Give slides and completed evaluation form to Director of Immunohistology or designee for evaluation. If sensitivity and specificity is optimal for the antibody the protocol that gave the best results can be entered into the protocol on the autostainer.
9. Antibody is added to antibody list.
10. File stained test slide with control slides in control slide cabinet.
11. File evaluation test sheet in new antibody logbook.